

Figure 1 (A-F)

Construct Forms Comprising at Least one Single-Stranded Region

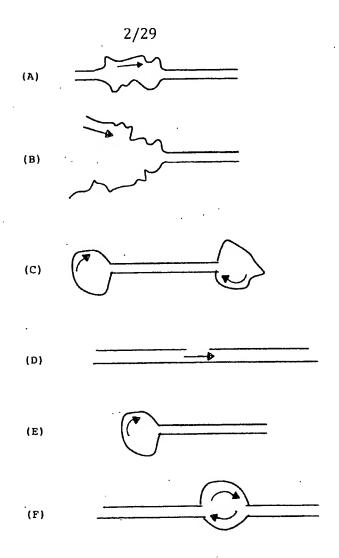
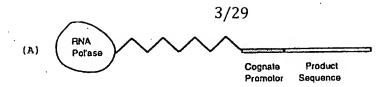


Figure 2 (A-F)

Functional Forms of the Construct



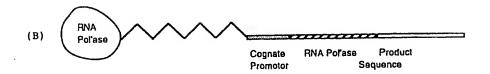




Figure 3 (A-C)

Three Constructs with an RNA Polymerase Covalently Attached to a Transcribing Cassette

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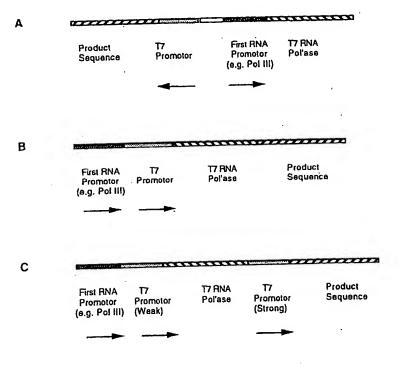


Figure 4 (A-C)

Three Constructs with Promoters for Endogenous RNA Polymerase

M13mp	M13mp18. Seq Length: 7250						
1.	AATGCTACTA	CTATTAGTAG	AATTGATGCC	ACCTTTTCAG	CTOCOCC		
51.	AAATGAAAAT	ATAGCTAÄAC	AGGITATTGA	CCATTTGCGA	AATGTATCTA		
101.	ATGGTCAAAC	TAAATCTACT	OGTTOGCAGA	ATTOGGAATC	AACTGTTACA		
151.	TGGAATGAAA	CTTOCAGACA	COGTACTITA	GTTGCATATT	TAAAACATGT		
201	TGAGCTACAG	CACCAGATTC	AGCAATTAAG	CTCTAAGCCA	TOOGCAAAAA		
251	TGACCTCTTA	TCAAAAGGAG	CAATTAAAGG	TACTCTCTAA	TOCTGACCTG		
301.	TTGGAGTTTG	CITCOGGICT	GGTTCGCTTT	GAAGCTOGAA	TTAAAACGCG		
351.	ATATTTGAAG	TCTTTCCCCCC	ттостсттаа.	TCTTTTTGAT	GCAATCCGCT		
401.	ПСПСТСА	CTATAATAGT	CAGGGTAAAG	ACCTGATTTT	TGATTTATGG		
451.	TCATTCTCGT	TTTCTGAACT	GTTTAAAGCA	TTTGAGGGGG	ATTCAATGAA		
501.	TATTTATGAC	GATTCCGCAG	TATTGGACGC	TATOCAGTCT	AAACATTTTA		
551.	CTATTACCCC	CTCTGGCAAA	ACTICITITIE	CAAAAGOCTC	TOGCTATTIT		
601.	GGTTTTTATC	GIOGICIOGI	AAAOGAGGGT	TATGATAGTG	TIGCTCTTAC		
651.	TATECCTOST	AATTCCTTTT	GOCGITATGT	ATCTGCATTA	GTTGAATGTG		
701.	GTATTCCTAA	ATCTCAACTG	ATGAATCTTT	CTACCTGTAA	TAATGTTGTT		
751.	COGITAGITC	GTTTTATTAA	CGTAGATTTT	TCTTCCCAAC	GTOCTGACTG		
801.	GTATAATGAG	CCAGTTCTTA	AAATCGCATA	AGGTAATTCA	CAATGATTAA		
851.	AGTTGAAATT	AAACCATCTC	AAGCCCAATT	TACTACTOGT	TCTGGTGTTC		
901.	TOGTCAGGGC	AAGCTTATT	CACTGAATGA	GCAGCITTGT	TACGITGATT		
951.	TOOGTAATGA	ATATOCGGTT	CTTGTCGAAG	ATTACTCTTG	ATGAAGGTCA		
1001	GOCAGOCTAT	COCOCTOCTIC	TGTACACCGT	TCATCTGTCC	TCTTTCAAAG		
1051	TTGGTCAGTT	CEGTICCCTT	ATGATTGACC	GICIGOGOCT	OGITICOGGCT		
1101	AAGTAACATG	GAGCAGGTOG	COGGATTTCCGA	CACAATTTAT	CAGGOGATGA		
1151	TACAAATCTC	CGTTGTACCTT	таптововс	TTGGTATAAT	OCCIGOGOGI		
1201	CAAAGATGAG	TGTTTTAGTG	TATTCTTTCG	CCICTICGI	TITAGGTTGG		

Figure 5

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1251	TGCCTTCGTA	GTGGCATTAC	GTATTTTACC	COTTTAATCG	AAACTTCCTC
1301	ATGAAAAAGT	CTTTAGTCCT	CAAAGOCTCT	GTAGOOGTTG	CTACCCTCGT
1351	TOOGATGCTG	TCTTTCGCTG	CTGAGGGTGA	OGATOCOGCA	AMAGOGGCCT
1401	TTAACTCCCT	GCAAGOCTCA	COCACOGAAT	ATATOGGTTA	TEOGTEGGC
1451	ATGGTTGTTG	TCATTGTOGG	COCAACTATC	GETATCAAGC	TGTTTAAGAA
1501	ATTCACCTCG	AAAGCAAGCT	GATAAACCGA	TACAATTAAA	CCTCCTTTT
1551	@GAGCCTTTT	TTTTTGGAGA	TTTTCAACGT	GAAAAAATTA	TTATTOGCAA
1601	TTCCTTTAGT	TGTTCCTTTC	TATTCTCACT	COCCTIGARAC	TGTTGAAAGT
1651	TGTTTAGCAA	AACCCCATAC	AGAAAATTCA	TTTACTAACG	TCTGGAAAGA
1701	CGACAAAACT	TTAGATOGTT	ACGCTAACTA	TGAGGGTTGT	CTGTGGAATG
1751	CTACAGGOGT	TGTAGTTTGT	- ACTEGTGACG	AAACTCAGTG	TTACGGTACA
1801	TGGGTTCCTA	песеспес	TATOCCTGAA	AATGAGGGTG	GTOGCTCTGA
1851	GGG GGCGGT	TOTGAGGGTG	GOOGTTCTGA	COCITO COCITO	ACTAAACCTC
1901	CTGAGTACGG	TGATACACCT	ATTOOGGGCT	ATACTTATAT	CAACCCTCTC
	GACCGCACTT	ATOCGOCTEG	TACTGAGCAA	AACCCGCTA	ATOCTAATOC
2001	TICTCTTGAG	GAGTCTCAGC	CTCTTAATAC	TITCATGTTT	CAGAATAATA
2051	GGTTCCGAAA	TAGGCAGGGG	GCATTAACTG	TITATACGGC	CACTGTTACT
2101	CAAGGCACTG	ACCCCGTTAA	AACTTATTAC	CAGTACACTC	CTGTATCATC
2151	AAAAGCCATG	TATGACGCTT	ACTEGAACEG	TAAATTCAGA	GACTGOGCTT
220		ACCCCGTTAA	AACTTATTAC	CAGTACACTC	CTGTATCATC
215	1 AAAAGOCATG	TGCCTCAACC	TOCTGTCAAT	ecleaceace	ECICIEGIEG
220	1 TOCATTICTIGG	CTTTAATCAA	GATOCATTOG	TTTGTGAATA	TCAAGGCCAA
225			TOCTGTCAAT	ecteeceece	ecticiestes
230			AGGGTGGTGG	CICTGAGGGT	GEOGETTCTG
235		CTCTGAGGGA	G G G G G G G G G G G G G G G G G G G	GIEGIEGCIC	TOGTICOGGI
240		/	GGCAAACGCT	AATAAGGGGG	CTATGACOGA
	1 AAATGOOGAT		TACAGTOTGA	COCTAMAGOC	AAACTTGATT
			Floring E		

Figure 5

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2501	CTGTCGCTAC	TGATTACGGT	GCTGCTATCG	ATGGTTTCAT	TGGTGAOGIT
2551	TOOGGOOTIG	CTAATGGTAA	TOGTOCTACT	GGTGATTTTG	CTGGCTCTAA
2601	TTCCCAAATG	OCTICANGTOG	GTGACCETGA	TAATTCACCT	TTAATGAATA
2651	ATTTCCGTCA	ATATTTACCT	TOOCTOOCTC	AATOGGTTGA	ATGTOGOCCT
2701	TTTGTCTTTA	COCCTOCATION	ACCATATGAA	TTTTCTATTG	ATTGTGACAA
2751	AATAAACTTA .	TTOOGTEGGTG	TCTTTGCGTT	TCTTTTATAT	GTTGCCACCT
2801	TTATGTATGT	ATTTTCTACG	TTTGCTAACA	TACTGCGTAA	TAAGGAGTCT
2851	TTATCATGCC	AGTTCTTTTG	GGTATTCCGT	TATTATTGCG	THOCIOGGI
2901	ПССПСТСС	TAACTITGIT	COCCTATCTG	CTTACTTTTC	TTAAAAAGGG
2951	CTTCGGTAAG	ATAGCTATTG	CTATTTCATT	GTTICTTGCT	CTTATTATTG
3001	GCCTTAACTC	AATTCTTGTG	G GTTATCTCT	CTGATATTAG	OGCTCAATTA
3051	COCTCTGACT	TIGTTCAGGG	TGTTCAGTTA	ATTICTCCCCGT	CTAATGOGCT
3101	TCCCTGTTTT	TATGTTATTC	TCTCTGTAAA	GCCTCCTATT	TTCATTTTTG
3151	ACGTTAÁACA	AAAAATCGTT	TCTTATTTGG	ATTGGGATAA	ATAATATGGC
3201	TGTTTATTTT	GTAACTGGCA	AATTAGGCTC	TOGAMAGACG	CTOGTTAGOG
3251	TTGGTAAGAT	TCAGGATAAA	ATTGTAGCTG	GGTGCAAAAT	AGCAACTAAT
3301	CTTGATTTAA	GCCTTCAAAA	OCTOCCOCAA	GTOGGGAGGT	TOGCTAAAAC
3351·	COCTOCCCTT	CTTAGAATAC	COGGATAAGCC	TTCTATATCT	GATTTGCTTG
3401	CTATTGGGGGG	COGTAATGAT	TOCTACGAATG	AAAATAAAAA	осеспест
3451	GITCTOGATG	AGTGCGGTAC	TTGGTTTAAT	ACCOGNICIT	GGAATGATAA
3501	CCAAAGACAG	COCATTATTG	ATTEGTTTCT	ACTOCTOGT	AAATTAGGAT
3551	GGGATATTAT	ттствт	CAGGACTTAT	CTATTGTTGA	TAMACAGGCG
3601	OGITICIGCAT	TAGCTGAACA	TGTTGTTTAT	TGTOGTOGTC	TOGACAGAAT
3651	TACTITACCT	TTTGTCGGTA	CTTTATATTC	TCTTATTACT	GCCTCGAAAA
3701	теостстеос	TAAATTACAT	eileccelle	TTAAATATGG	CGATTCTCAA
3751	TTAAGCCCTA	CTGTTGAGCG	TTGGCTTTAT	ACTOGTAAGA	ATTTGTATAA
3801	OGCATATGAT	ACTAMACAGG	CTTTTCTAG	TAATTATGAT	1000 दादारा

Figure 5

3851	ATTCTTATTT	AACGCCTTAT	TTATCACAOG	GIOGGIATIT	CAAAOCATTA
3901	AATTTAGGTC	AGAAGATGAA	ATTAACTAAA	ATAATATTGA	AAAAGTTTTC
3951	TOSOGTICIT	TGTCTTGCGA	TTGGATTTGC	ATCAGCATTT	ACATATAGTT
4001	ATATAACCCA	ACCTAAGCCG	GAGGITAAAA	AGGTAGTCTC	TCAGACCTAT
4051	GATTTTGATA	AATTCACTAT	TGACTCTTCT	CAGOGTOTTA	ATCTAAGCTA
4101	TOGOTATGTT	TTCAAGGATT	CTAAGGGAAA	TAATTAATTA	AGOGACGATT
4 1'5 1	TACAGAAGCA	AGGTTATTCA	CTCACATATA	TTGATTTATG	TACTGTTTCC
4201	ATTAAAAAAG	GTAATTCAAA	TGAAATTGTT	AAATGTAATT	AATTTIGITT
4251	TCTTGATGTT	TGTTTCATCA	тсптспппа	CTCAGGTAAT	TGAAATGAAT
4301	AATTOGOCTC	TGCGCGATTT	TGTAACTTGG	TATTCAAAGC	AATCAGGGGA
4351	AATCCGTTATT	GITTCICCCCG	ATGTAAAAGG	TACTGTTACT	GTATATTCAT
4401	CTGAOGTTAA	ACCTGAAAAT	CTACGCAATT	TCTTTATTTC	TGTTTTACGT
4451	GCTAATAATT	TTGATAATGGT	TEGITCAATT	CCTTCCATAA	TTCAGAAGTA
4501	TAATOCAAAC	- AATCAGGATT	ATATTGATGA	ATTGCCATCA	TCTGATAATC
4551	AGGAATATGA	TGATAATTCC	ecicciicis	GIGGITICIT	TGTTCCGCAA
4601	AATĢATAATG	TTACTCAAAC	TTTAAAATT	AATAAOGTTC	GGGCAAAGGA
4651	TTTAATACGA	GTTGTCGAAT	TGTTTGTAAA	GTCTAATACT	TCTAAATCCT
4701	CAAATGTATT	ATCTATTGAC	GECTETAATE	TATTAGTTGT	TAGTECTECT
4751	AAAGATATTT	TAGATAACCT	TOCTCAATTC	CTTTCTACTG	TTGATTTGCC
4801	AACTGACCAG	ATATTGATTG	AGGGTTTGAT	ATTTGAGGTT	CAGCAAGGTG
4851	ATGCTTTAGA	TTTTCATTT	ectecteect	CTCAGOGTGG	CACTGTTGCA
4901	GEOGRATIA	ATACTGACCG	OCTICACCTICT	GTTTATCTT	CIECTEGIEG
4951	TICGTICGGT	ATTTTTAATG	GOGATGTTTT	AGGGCTATCA	GTT0303CAT
5001	TAAAGACTAA	TAGOCATTCA	AAAATATTGT	CTGTGCCACG	TATTCTTACG
5051	CTTTCAGGTC	AGAAGGGTTC	TATCTCTGTT	GGCCAGAATG	TOCCTTTTAT
5101	TAAAGACTAA	TAGOCATTCA	AAAATATTGT	CTGTGCCACG	TATTCTTACG
5151	OGATTGAGOG	TCAAAATGTA	GGTATTTCCA	TGAGOGTTTT	TOCTGTTGCA

-Figure 5

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5201	ATGGCTGGGG,	GTAATATTGT.	TCTGGATATT	ACCAGCAAGG	COGATAGITT
5251	GAGITICTCT	ACTCAGGCAA	GIGATGTTAT	TACTAATCAA	AGAAGTATTG
5.301	CTACAACGGT	TAATTTGCGT	GATGGACAGA	CTCTTTTACT	COGTOCOCTC
5351	ACTGATTATA	AAAACACTTC	TCAAGATTCT	GGOGTACOGT	TOCTGTCTAA
5401	AATCCCTTTA	ATCCCCCTCC	TGTTTAGCTC	COSCTCTGAT	TOCAACGAGG
5451	AMAGCACGTT	ATAOGTGCTC	GTCAAAGCAA	CCATAGTACG	OCCOCTICTAG
5501	CCCCCCATTA	ACCICICATION	GIGIGGIGGI	TACGCGCAGC	GTGACCGCTA
5551	CACTTECCAG	COCCTAGOG		TOGOTHOT	σοσποσιτί
5601	CTOGOCAOGIT	TOGOCGGCTT	TOOCOGTCAA	GCTCTAAATC	GEGEGETTOCC
5651	TTTAGGGTTC	CGATTTAGTG	CTTTACCGCCA	OCTOGACCCC	AAAAAACTTG
5701	ATTTGGGTGA	TEGTTCACGT	AGTGGGCCAT	OCCOCTGATA	GACGGTTTTT
5751	COCCTTTGA	COTTOGAGTO	CACGITCITT	AATAGTGGAC	TCTTGTTCCA
5801	AACTGGAACA	ACACTCAACC	CTATCTOGGG	CTATTCTTTT	GATTTATAAG
5851	GGATTTTGCC	GATTTOGGAA	CCACCATCAA	ACAGGATTTT	COCCTRICTION
5901	GGCAAACCAG	OGTIGGACOGC	TTECTECAAC	TCTCTCAGGG	CCAGGOGGTG
5951	AAGGGCAATC	AGCTGTTGCC	OGICIOGCIG	GTGAAAAGAA	AAAOCAOOCT
6001	GGCGCCCAAT	ACCICAMACICG	CTCTCCCCCCG	COCCITICACC	GATTCATTAA
6051	TECACCTECC	ACCACAGGIT	TOOOGACTEG	AAAGOOGGCA	GTGAGOGCAA
6101	CGCAATTAAT	GTGAGTTAGC	TCACTCATTA	CCCACCCCAG	GCTTTACACT
6151	TTATGCTTCC	GECTOGTATG	TIGIGIGGAA	TIGIGAGOGG	ATAACAATTT
6201	CACACAGGAA	ACAGCTATGA	CCATGATTAC	GAATTOGAGC	TOGGTACCOG
6251	GOGATOCTCT	AGAGTOGACC	TECAGECATE	CAAGCTTGGC	ACTEGEOGTC
6301	GTTTTACAAC	GTOGTGACTG	GGAAAACCCT	. GEOGTTACCC	AACTTAATCG
6351	CTTGCAGCA	CAATCCCCTT	TOGOCAGCTG	GOGTAATAGC	GAAGAGGCCC
640	CACCGATCG	COCTTOCCAA	CÁGITGOGCA	COCTGAATEG	OGAATGGCGC
645	THEOCIGGI	TTCCCCCACC	AGAAGOOGTG	COGGAAAGCT	COCTOCACTG
650			OGGIOGIOGI	COCCTCAAAC	TEGCAGATEC
					•

Figure 5

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6551	ACCITTACGA	TEXECUTATE	TACACCAACG	TAAOCTATOC	CATTACGGTC
6601	AATOOGOOGT	TIGTTCCCAC	CCACAATTOOG	ACGGGTTGTT	ACTOGCTCAC
6651	ATTTAATGTT	GATGAAAGCT	GOCTACAGGA	ACCOCAGACG	CGAATTATTT
67 [.] 01	TIGATGGCGT	TOCTATTGGT	TAAAAAATGA	GCTGATTTAA	CAAAAATTTA
6751	ACGCGAATTT	TAACAAAATA	TTAACGTTTA	CAATTTAAAT	ATTIGCTTAT
6801	ACAATCTTCC	TGTTTTGGG	COTTTCTGA	TTATCAACCG	GGGTACATAT
6851	GATTGACATG	CTAGTTTTAC	GATTACCGTT	CATCGATTCT	спаттест
6901	CAGACTCTC	AGGICAATIGAC	CTGATAGOCT	TTGTAGATCT	CTCAAAAATA
6951	GCTACCCTCT	COGGCATGAA	TTTATCAGCT	AGAACGGTTG	AATATCATAT
7001	TGATGGTGAT	TIGACIGICT	COCCOCTTTC	TCACCCTTTT	GAATCTTTAC
7051	CTACACATTA	CTCAGGCATT	GCATTTAAAA	TATATGAGGG	TTCTAAAAAT
7101	TTTTATCCTT	COGTTGAAAT	AMAGGCTTCT	CCCCCAAAAG	TATTACAGGG
7151	TCATAATGTT	TTTGGTACAA	COGATTTAGC	TTTATGCTCT	GAGGCTTTAT

Figure 5

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COMPLEMENTARY TO M₁₃

	•		
POSITION 6 4 5	5 ' 3' AGCAACACTATCATÀ	POSITION 631	M ₁₃ /1
615	ACGACGATAAAAACC	601	M ₁₃ /2
585	TTTTGCAAAAGAAGT	571	M ₁₃ /3
555	AATAGTAAAATGTTT	541	M ₁₃ /4
525	CAATACTGCGGAATG	511	M ₁₃ /5
495	TGAATCCCCCTCAAA	481	M ₁₃ /6
465	AGAAAACGAGAATGA	451	M ₁₃ /7
435	CAGGTCTTTACCCTG	. 421	M ₁₃ /8
405	AGGAAAGOGGATTGC	391	M ₁₃ /9
375	AGGAAGOOOGAAAGA	361	M ₁₃ /10

COMPLEMENTARY TO SS PHAGE DNA

POSITION		POSITION	
351	5' 3' ATATITGAAGTCTTT	366	M ₁₃ /11
371	TCTTTTTGATGCAAT	386	M ₁₃ /12
391	CTATAATACTCAGGG	406	M ₁₃ /13
411	TGATTTATGGTCATT	426	· M ₁₃ /14
431	GTTTAAAGCATTTGA	446	M ₁₃ /15
451	TATTTATGACGATTC	466	M ₁₃ /16
471	TATOCAGTCTAAACA	486	M ₁₃ /17
491	CTCTGGCAAAACTTC	506	M ₁₃ /18
511	TOGOTATTTTGGTTT	526	M ₁₃ /19
-531	AAACGAGGGTTATGA	546	M _{13/2} 0

Figure 6

Primers for Nucleic Acid Production Derived from M13mp18 Sequence

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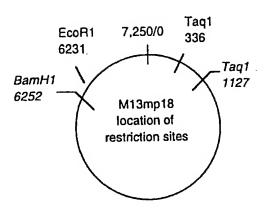


Figure 7

Appropriate M13mp18 Restriction Sites

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Lane 1: from calf thymus + Taq digested mp18 amplification reaction

Lane 2: from Taq digested mp18 amplification reaction

Lane 3: from calf thymus amplification reaction

Lane 4: øX174 Hinf1 size marker

Figure 8

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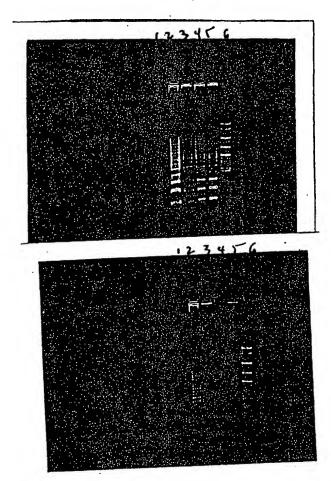
Lane 1: no template

Lane 2: mp18 template, phosphate buffer

Lane 3: Mspl/pBR322 size marker Lane 4: mp18 template, MOPS buffer

Figure 9

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Top= (+) Template
Bottom= (-) Template

Lane 1: phosphate buffer

Lane 2: MES Lane 3: MOPS Lane 4: DMAB Lane 5: DMG

Lane 6: pBR322/Mspl size marker

Figure 10

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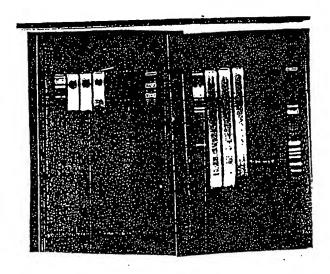
Lane 1: DMAB buffer, no template Lane 2: DMAB buffer, mp18 template Lane 3: DMG buffer, no template Lane 4: DMG buffer, mp18 template

Lane 5: No reaction

Lane 6: 200 ng Taq I digested mp18 size marker/positive control

Figure 11

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First Time Interval Second Time Interval

Agarose Gel Analysis

Lane 1: lambda Hind III marker

Lane 2: Amp/Untreated

Lane 3: Amp/Kinased

Lane 4: Amp/Kinased/Ligated

Lane 5: PCR/Untreated

Lane 6: PCR/Kinased

Lane 7: PCR/Kinased/Ligated

Lane 8: øX174/Hinf1 marker

Figure 12 ·

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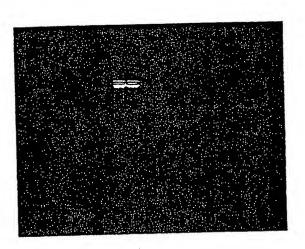
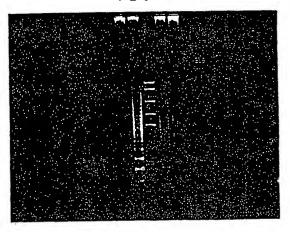


Figure 13

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1 2 3 4 5 6



Lane 1: Primers alone

Lane 2: Primers + taq digested M13 DNA

Lane 3: Molecular weight markers

Lane 4: Primers + RNA

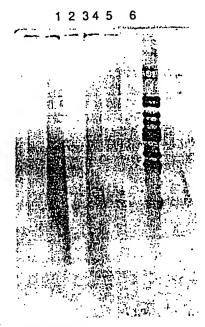
Lane 5: Primers alone

Lane 6: M13 digested DNA

Buffer was dimethyl amino glycine, pH 8.6

Figure 14

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Lane 1: Primers alone

Lane 2: Primers + taq digested M13 DNA

Lane 3: Molecular weight markers

Lane 4: Primers + RNA
Lane 5: Primers alone

Lane 6: M13 digested DNA

Buffer was dimethyl amino glycine, pH 8.6

Figure 15



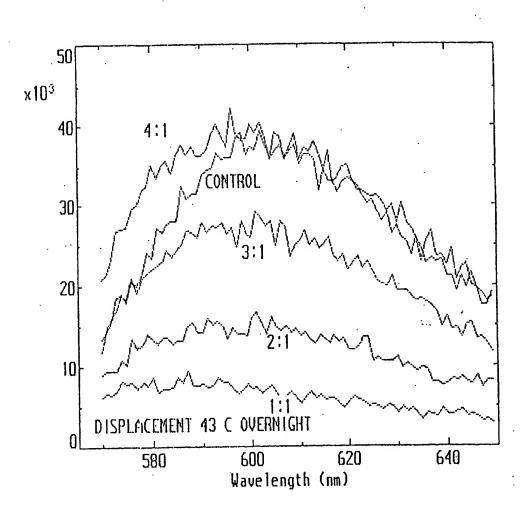


Figure 16

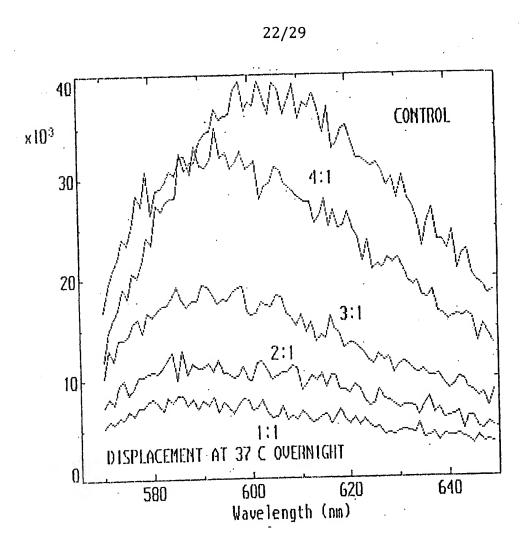


Figure 17

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piBI 31-BH5-2

fmet AUG of Lac z [T7 Promotor region---LAC PROMOTOR.ATG ACC ATG ATT ACG CCA GAT ATC AAA TTA ATA CGA CTC ACT ATA

oligo 50-mer 3'- tac t'aa t'gc ggt' ct'a t'ag t'Vt aat' tat' gct' gag t'ga t'at' c-5'

T7 RNA Start {«« T3 Promotor Region }
IGGG CTC ICCT TTA GTG ACG GTT AAT
...»»} «- T3 Start Signal

pIBI 31 BSII/HCV

("- T7 Promotor Region)

MULTIPLE CLONING SITE + 390 BASE INSERT CTA /TAG TGA GTC CGT ATT AAT....

"- T7 Start Signal

5'-ct'a t'ag t'ga gt'c gt'a tt'a at'...........

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 		 	3'
 	 	 ~~~~~~	

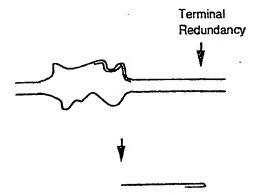
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Replication Bubble with Nucleotide Analogs



Primer-Dependent DNA Production Using Nucleic Acid Construct

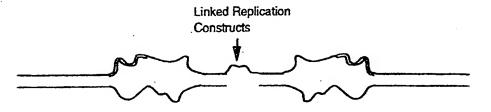
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Hairpin Product

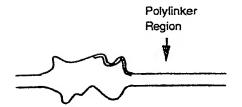
Figure 21

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Linked Complementary Production Constructs

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Cloning Site in Production Constructs

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### ARRANGEMENT OF OLIGONUCLEOTIDE PRIMERS IN AMPLIFICATION REACTION

1	2	3	4	5	6	7	8	9	10
20	19	18	17	16	15	14	13	12	11